British Heart Journal, 1978, **40**, 526-529

Platelet adhesiveness, plasma fibrinogen, and fibrinolytic activity in acute myocardial infarction

SUBHASH C. SHARMA AND H. N. SETH

From Army Hospital, Delhi Cantt.-110010, India

SUMMARY In a search for a possible 'hypercoagulable' state in myocardial infarction, platelet adhesiveness, plasma fibrinogen, and euglobulin lysis time were estimated in 48 cases of acute myocardial infarction and in 15 age-matched normals. An increase in platelet adhesiveness ($P<0.05$), plasma fibrinogen ($P<0.01$), and fibrinolytic activity ($P<0.001$) was noted in the patients when compared with normals. Our data suggest that there are no obvious disturbances in the dynamic equilibrium normally existing between coagulation and fibronolysis as postulated by Astrup (1956) in patients with acute myocardial infarction.

Ischaemic heart disease represents the commonest serious health problem of contemporary society and acute myocardial infarction remains the main cause of in-hospital deaths (Braunwald, 1976). Despite great advances made recently in the understanding of the pathophysiology of coronary heart disease, the aetiology still remains unclear. Though atherosclerotic coronary disease is the principal aetiological factor in ischaemic heart disease, the role of thrombus formation and a state of hypercoagulability in relation to pathogenesis are not fully understood. In 1852 Rokitansky proposed that atherosclerotic plaques resulted from the organisation of thrombi. Virchow (1863), however, disputed this claim; but recently attention has been again drawn to thrombus, mainly because of its components, namely platelets and fibrin. Platelets play a pivotal role in coagulation and thrombogenesis. Variable platelet adhesiveness in ischaemic heart disease has been reported (Genton and Steele, 1975). It has also been suggested that the increase in plasma fibrinogen may predispose to thrombus formation by increasing the blood viscosity (Wells and Merrill, 1961; Burch and dePasquale, 1962). Hence it is logical to assume that altered fibrinolytic activity in such susceptible individuals is likely to play an important role in the final outcome.

Review of the published material indicates some measure of dispute (Røjel, 1959; Sikka *et al.*, 1967; Chandler *et al.*, 1974). The purpose of this investigation is to assess platelet adhesiveness, plasma fibrinogen, and fibrinolytic activity in cases of acute myocardial infarction and to compare patients with age-matched normals.

Received for publication 18 April 1977

Subjects and methods

Studies were made on 63 subjects, 48 of whom were in Group A, and 15 in Group B.

Group A: 48 cases of acute myocardial infarction, 26 to 85 years of age, were admitted to this hospital and diagnosis was based upon typical history, electrocardiographic findings, and enzyme studies (Lawrie *et al.*, 1967). Cases of diabetes mellitus and hypertension with acute myocardial infarction were excluded from the study.

Group B: 15 normal healthy subjects, 25 to 80 years of age acted as controls.

Platelet adhesiveness was estimated by the method of Eastham (1964) using adenosine diphosphate. Since many factors such as diet (Philp and Wright, 1965), physical exertion (Pegrum *et al.*, 1967), and smoking (Ashby *et al.*, 1965) affect platelet adhesiveness, the procedure was standardised as rigidly as was practicable. A team of medical officers and laboratory staff specially designated for such cases was available on call round the clock throughout this study. Blood was collected in siliconised syringes from group A immediately on admission. Blood samples were collected within 2 hours of the appearance of the first clinical symptoms in all patients. Blood from group B was collected after an overnight fast on 2 consecutive days. Each subject abstained from smoking for at least 3 hours before the venepuncture. Physical activity was reduced to a minimum in all the subjects. The method for

estimation of platelet adhesiveness was as follows: 2 ml blood was added to 2·4 mg ethylenediaminetetra acetic acid (EDTA) and to 2 mg heparin in separate polystyrene bottles and mixed in a Matburn mixer at 30 rev/min. To the heparinised sample 0·04 ml ADP solution was added (from a stock solution of 5 mg in 20 ml saline) and the bottles were returned to the mixer. After exactly 30 minutes the ADP treated sample was transferred to another polystyrene bottle containing 2·4 mg EDTA. Samples were mixed for 30 minutes and platelet counts were performed as soon as possible but in all cases within an hour of collection. Counts were performed in duplicate using the improved Neubaur chamber and the results were averaged. The platelet adhesiveness was expressed as a percentage as follows:

$$\frac{\text{Total EDTA sample count—heparinised sample ADP count}}{\text{Total EDTA sample count}} \times 100$$

Plasma fibrinogen was estimated by the method of Ratnoff and Menzie (1951) and euglobulin lysis time by the method of Cash (1966). Patients were not given any drug known to interfere with above estimations. Statistical analysis was carried out by Student's t test.

Results

Age, platelet adhesiveness, plasma fibrinogen, and euglobulin lysis time in respect of each group are given in the Table. Mean platelet adhesiveness in groups A and B was 65·5 per cent and 61·5 per cent, respectively, and the difference was statistically significant ($P < 0.05$). The total platelet counts in the two groups were essentially similar. Plasma fibrinogen was significantly more ($P < 0.01$) in patients when compared with normal subjects. Euglobulin lysis time was conspicuously diminished in the patients and the difference between the groups was highly significant ($P < 0.001$).

Table *Platelet adhesiveness (PA), plasma fibrinogen, and euglobulin lysis time (ELT) in acute myocardial infarction (group A) and normals (group B): values are mean ± SD, n is number of subjects*

	Mean age (y) (range)	*PA (%)*	*Fibrinogen (mg/100 ml)*	*ELT (min)*
Group A	54·9	65·5	321·9	370·7
(n 48)	(26–85)	±8·1	±180·4	±59·1
Group B	50·3	61·5	253·4	307·7
(n 15)	(25–80)	±8·9	±37·0	±31·6
P value				
A vs B	—	<0·05	<0·01	<0·001

Discussion

It is now established that transmural infarction is associated with a completely occlusive lesion in one or more coronary arteries, most commonly the artery that supplies the infarcted area (Chandler, 1974; Chandler *et al.*, 1974). These cases have one feature in common; that is severe stenosing coronary atherosclerosis. In recent years there has been an attempt to explain this combination of findings on the basis of primary myocardial ischaemia caused by some exacerbation of the stenosing process (Spain and Bradess, 1960; Roberts and Buja, 1972; Roberts, 1974) or to a mechanism or mechanisms independent of events in the main coronary circulation (Anderson, 1970; Baroldi, 1972).

PLATELET ADHESIVENESS

The adhesion of platelets to the injured endothelium in the vessel wall is the first step in the formation of a thrombus. Hughes and Tonks (1956, 1959, 1962, 1968) were probably the first to study platelet emboli and autologous whole blood emboli in the myocardial circulation. Their experiments showed clear evidence of vasculitis and tissue damage from platelet aggregate emboli. What initiates the process is not clear but the tendency to platelet adhesion/aggregation appears to be an important link in the sequence of changes in the circulating blood contributing to the development of thrombosis, especially of arterial thrombosis. Extensive interest has been focussed recently on platelet adhesiveness in various diseases (Seth, 1973; Steele *et al.*, 1973; Sharma *et al.*, 1977). Despite descrepancies in results presented by different authors there is evidence that high platelet adhesiveness predisposes to thrombosis. High platelet adhesiveness noted in this study is in agreement with majority of other reports.

PLASMA FIBRINOGEN

Our data show a significant ($P < 0.01$) increase in plasma fibrinogen in patients with acute myocardial infarction as compared with normals. It has been suggested that the high fibrinogen in the plasma will greatly increase the viscosity of blood and predispose to thrombus formation (Wells and Merrill, 1961; Burch and dePasquale, 1962). It is known that the increase in plasma fibrinogen in the patients with coronary thrombosis is not transient in nature and, therefore, is to be distinguished from the short lived increase found in stress or tissue injury, such as burns, trauma, and infarction, and is probably the result of increased biosynthesis and an enhanced rate of turnover (Pilgeram *et al.*, 1973).

Whether the enhanced plasma fibrinogen results in the generation of potentially occlusive intravascular deposits of fibrin platelet thrombi will depend upon the balance between the rate of formation of fibrin and the rate of its metabolism or lysis.

EUGLOBULIN LYSIS TIME

Despite the criticism of substrate variability, the euglobulin lysis time is perhaps the best available method of assay of circulating plasminogen activator and hence a reliable indicator of fibrinolytic activity (Fletcher *et al.*, 1964; Fearnley, 1965). Our data reveal a significant ($P < 0.001$) increase of fibrinolysis in the patients as compared with the normals. However, the role of the fibrinolytic system in the pathogenesis as well as in the recovery stage of thromboembolic disorders is not yet clearly understood.

Thus a dynamic equilibrium as suggested by Astrup (1956) between coagulation and fibrinolysis, and a significant increase in platelet adhesiveness and fibrinogen with a corresponding decrease in euglobin lysis time in our patients, points towards a normally functioning and balanced state of equilibrium. It is, however, impossible to state whether these alterations are the result or are related to the cause of the event.

The authors are grateful to the Director General, Armed Forces Medical Services, for permission to publish this paper.

References

Anderson, T. W. (1970). Role of myocardium in the modern epidemic of ischaemic heart disease. *Lancet*, **2**, 753–755.

Ashby, P., Dalby, A. M., and Millar, J. H. D. (1965). Smoking and platelet stickiness. *Lancet*, **2**, 158–159.

Astrup, T. (1956). Biological significance of fibrinolysis. *Lancet*, **2**, 565–568.

Baroldi, G. (1972). Human myocardial infarction: coronarogenic or noncoronarogenic coagulation necrosis? In *Myocardiology, Recent Advances in Studies on Cardiac Structure and Metabolism*, Vol. 1, p. 399. Ed. by E. Bajusz and G. Rona. University Park Press, Baltimore.

Braunwald, E. (1976). Introductory remarks: protection of the ischemic myocardium. *Circulation*, **53**, Suppl. I, 1–2.

Burch, G. E., and De Pasquale, N. D. (1962). Editorial. Hematocrit, blood viscosity and myocardial infarction. *American Journal of Medicine*, **32**, 161–163.

Cash, J. D. (1966). Effect of moderate exercise on the fibrinolytic system in normal young men and women. *British Medical Journal*, **2**, 502–506.

Chandler, A. B. (1974). Mechanisms and frequency of thrombosis in the coronary circulation. *Thrombosis Research*, **4**, Suppl. I, 3–23.

Chandler, A. B., Chapman, I., Erhardt, L. R., Roberts, W. C., Schwartz, C. J., Sinapius, D., Spain, D. M., Sherry, S., Ness, P. M., and Simon, T. L. (1974). Coronary thrombosis in myocardial infarction. *American Journal of Cardiology*, **34**, 823–833.

Eastham, R. D. (1964). Rapid adhesive platelet count in whole blood. *Journal of Clinical Pathology*, **17**, 45–46.

Fearnley, G. R. (1965). *Fibrinolysis*, p. 35. Arnold, London.

Fletcher, A. P., Biederman, O., Moore, D., Alkjaersig, N., and Sherry, S. (1964). Abnormal plasminogen—plasmin system activity (fibrinolysis) in patients with hepatic cirrhosis: its cause and consequences. *Journal of Clinical Investigation*, **43**, 681–695.

Genton, E., and Steele, P. P. (1975). Platelet studies in patients with venous and cardiac disease. *Thrombosis et Diathesis Haemorrhagica*, **40**, Suppl., 223–226.

Hughes, A., and Tonks, R. S. (1956). Experimental embolic carditis. *Journal of Pathology and Bacteriology*, **72**, 497–503.

Hughes, A., and Tonks, R. S. (1959). The role of microemboli in the production of carditis in hypersensitivity experiments. *Journal of Pathology and Bacteriology*, **77**, 207–217.

Hughes, A., and Tonks, R. S. (1962). Intravascular platelet clumping in rabbits. *Journal of Pathology and Bacteriology*, **84**, 379–390.

Hughes, A., and Tonks, R. S. (1968). Lung and heart lesions from intravascular platelet clumping and its sequelae. *Journal of Pathology and Bacteriology*, **95**, 523–526.

Lawrie, D. M., Greenwood, T. W., Goddard, M., Harvey, A. C., Donald, K. W., Julian, D. G., and Oliver, M. F. (1967). A coronary-care unit in the routine management of acute myocardial infarction. *Lancet*, **2**, 109–114.

Pegrum, G. D., Harrison, K. M., Shaw, S., Haselton, A., and Wolff, S. (1967). Effect of prolonged exercise on platelet adhesiveness. *Nature (London)*, **213**, 301–302.

Philp, R. B., and Wright, H. P. (1965). Effect of adenosine on platelet adhesiveness in fasting and lipaemic bloods. *Lancet*, **2**, 208–209.

Pilgeram, L. O., Chee, A. N., and Von Dem Bussche, G. (1973). Evidence of abnormalities in clotting and thrombolysis as risk factors for stroke. *Stroke*, **4**, 643–657.

Ratnoff, O. D., and Menzie, C. (1951). A new method for the determination of fibrinogen in small samples of plasma. *Journal of Laboratory and Clinical Medicine*, **37**, 316–320.

Roberts, W. C., and Buja, L. M. (1972). The frequency and significance of coronary arterial thrombi and other observations in fatal acute myocardial infarction. A study of 107 necropsy patients. *American Journal of Medicine*, **52**, 425–444.

Roberts, W. C. (1974). Coronary thrombosis and fatal myocardial ischemia (Editorial). *Circulation*, **49**, 1–3.

Rokitansky, C. von (1852). *A Manual of Pathological Anatomy*. English translation London, Sydenham Society, 1852, pp. 261–272.

Røjel, J. (1959). A study of fibrinolysin activity in thrombotic diseases. *Acta Medica Scandinavica*, **164**, 81–93.

Seth, H. N. (1973). Fibrinolytic response to moderate exercise and platelet adhesiveness in diabetes mellitus. *Acta Diabetologica Latina*, **10**, 306–314.

Sharma, S. C., Vijayan, G. P., Suri, M. L., and Seth, H. N. (1977). Platelet adhesiveness in young patients with ischaemic stroke. *Journal of Clinical Pathology*, **30**, 649–652.

Sikka, K. K., Nath, K., Samuel, K. C., Gahlaut, D. S., Srivastava, M. C., and Gaffar, A. (1967). Plasma fibrinolytic activity in diabetes mellitus. *Journal of the Association of Physicians of India*, **15**, 279–282.

Spain, D. M., and Bradess, V. A. (1960). The relationship of coronary thrombosis to coronary atherosclerosis and ischemic heart disease. (A necropsy study covering a period of 25 years.) *American Journal of the Medical Sciences*, **240**, 701–710.

Steele, P. P., Weily, H. S., Davies, H., and Genton, E. (1973). Platelet function studies in coronary artery disease. *Circulation*, **48**, 1194–1200.

Virchow, R. (1863). *Cellular Pathology. As Based upon Physiological and Pathological Histology*. Translated from the second German edition originally published by J. B. Lippincott, New York, Dover Publications, 1971.

Wells, R. E., and Merrill, E. W. (1961). Editorial. The variability of blood viscosity. *American Journal of Medicine*, **31**, 505–509.

Requests for reprints to Major Subhash C. Sharma, Department of Pathology, All India Institute of Medical Sciences, New Delhi-110016, India